LOCAL ELECTROCORTICAL RESPONSES INDUCED BY TOPICAL APPLICATION OF ACETYLCHOLINE CHLORIDE TO HUMAN CEREBRAL CORTEX*

JOSEPH G. CHUSID and C. G. DE GUTIERREZ-MAHONEY

The Neurological Division, St. Vincent's Hospital, New York

STUDIES dealing with the electrocortical effects following topical application of acetylcholine chloride solutions to the cerebral cortex of animals have been reported previously.[1-9] The present investigation was undertaken in an attempt to determine for man the patterns of cerebral electrocortical response to acetylcholine chloride solutions similarly tested in a group of patients undergoing cerebral operations.

Method: Acetylcholine chloride solutions were freshly prepared at the time of operation in concentrations of 10 per cent or 20 per cent in physiological saline. Small cotton pledgets (1.5 mm. squares) were saturated with test solution and applied to exposed cerebral cortex for varying periods of time. In most instances, electrocorticograms were made with the pledgets in situ and with the aid of Marshall cortical electrodes and a Type III Grass six channel electroencephalograph machine. Cortex to cortex tracings were commonly employed with an interelectrode distance ranging from 0.8 to 5 centimeters. Some tracings were also made following the brief application to the cerebral cortex of small filter paper squares soaked in freshly prepared 2 per cent strychnine sulfate solution. All patients received Demerol,® morphine sulfate and atropine sulfate or scopolamine 1½ to 3 hours preoperatively and operation was usually performed under general anesthesia (nitrous oxide, pentothal-nitrous oxide, avertin-nitrous oxide).

Results: In six of the twelve patients tested, a profound change in the local electrocorticogram occurred after the topical application of 10 per cent or 20 per cent acetylcholine chloride solutions (Table I). In the twenty-nine tests made, a severe change in the electrocorticogram

* Presented February 19, 1952 before the Combined Meeting of The New York Neurological Society and Section of Neurology and Psychiatry at The New York Academy of Medicine.
Manuscript received for publication February, 1952.

TABLE I — INDICATING RESULTS FOLLOWING TOPICAL APPLICATION OF ACETYLCHOLINE CHLORIDE TO HUMAN CEREBRAL CORTEX

Case	*Clinical Diagnosis*	*Preoperative Medication*	*Anesthesia*	*Area of Application*	*Max. Duration of Application*	*No. of Applications*		*"Positive" Reactions*
						10%	*20%*	
(1) M.P.	Right Frontal Glioblastoma	Sodium Luminal & Atropine Sulfate	Nitrous oxide	Right Antero-Lateral Frontal	2 mins.	2	0	2
(2) R.B.	Left Frontal Metastatic Carcinoma	Atropine Sulfate	Pentothal & Nitrous oxide	Left Lateral Frontal	12 mins.	6	0	2
(3) J.R.	Multiple Metastatic Hypernephroma	Morphine Sulfate & Atropine Sulfate	Pentothal & Nitrous oxide	Left Lateral Frontal	17 mins.	2	0	1
(4) E.G.	Left Frontal Metastatic Carcinoma	Morphine Sulfate & Scopolamine	Local	Bilateral Posterior Parietal	24 mins.	0	4	1
(5) O.B.	Left Middle Cerebral Thrombosis	Demerol & Atropine Sulfate	Pentothal & Nitrous oxide	Bilateral Posterior Parietal	63 mins.	1	4	0
(6) R.G.	Psychomotor Epilepsy	Morphine Sulfate & Scopolamine	Nitrous oxide & Pentothal	Right Lateral Frontal	31 mins.	0	1	0
(7) S.K.	Left Cerebral Metastatic Carcinoma	Demerol & Atropine Sulfate	Nitrous oxide & Pentothal	Right Medial Frontal	10 mins.	1	2	0
(8) F.P.	Left Middle Cerebral Thrombosis	Morphine Sulfate & Atropine Sulfate	Avertin & Nitrous oxide	Right Medial Frontal	11½ mins.	0	1	1
(9) V.E.	Left Parietal Metastatic Carcinoma	None	Avertin & Nitrous oxide	Right Medial Frontal	27 mins.	0	1	0
(10) M.A.	Traumatic Encephalopathy	Demerol & Atropine Sulfate	Avertin & Nitrous oxide	Right Medial Frontal	45 mins.	0	2	0
(11) E.E.	Congenital Communicating Hydrocephalus	Demerol & Scopolamine	Avertin & Nitrous oxide	Right Medial Frontal	12 mins.	0	1	1
(12) A.E.	Focal Epilepsy	Demerol & Scopolamine	Pentothal & Nitrous oxide	Right Lateral Parietal	28 mins.	1	0	0

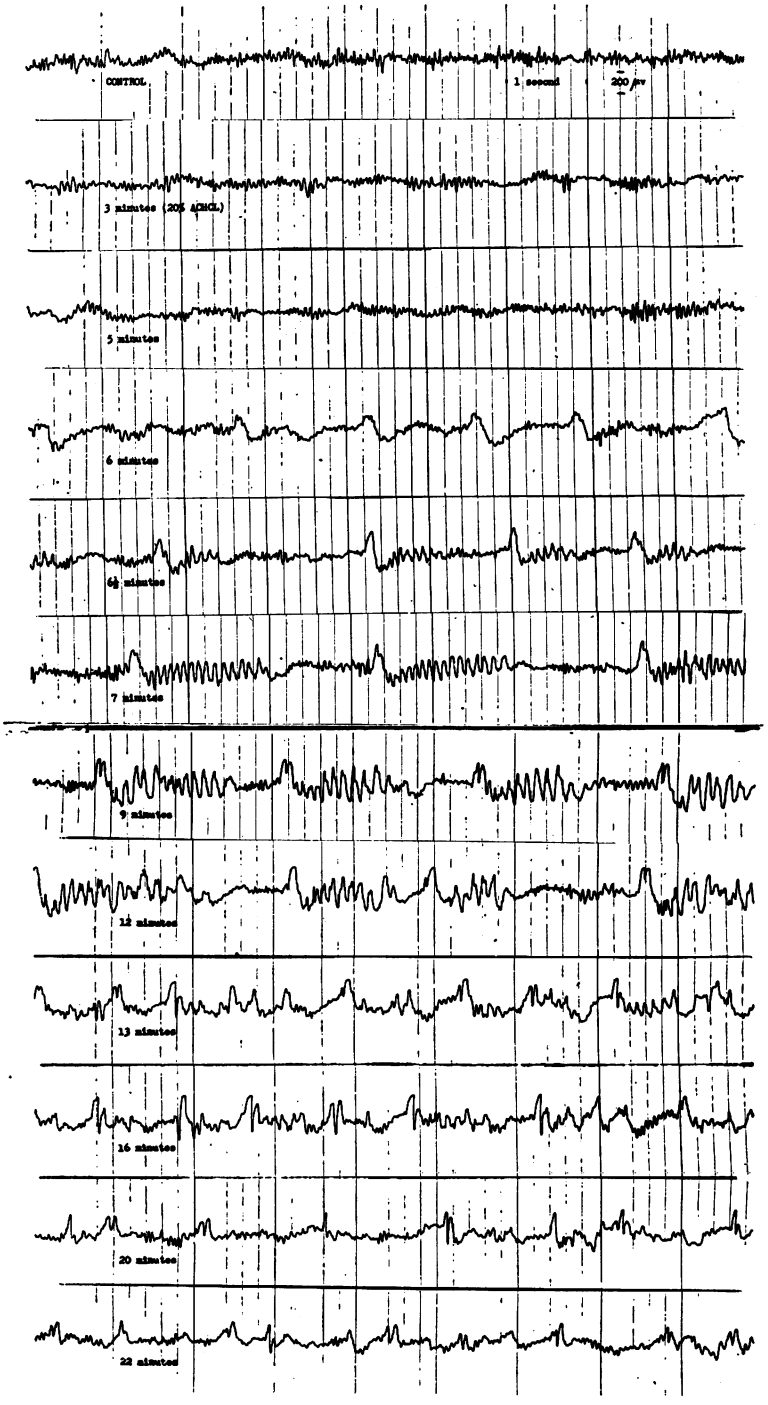

FIGURE 1—Serial electrocorticograms showing the local effects after application of 20% acetylcholine chloride for twelve minutes to a right frontal cortical area (Case 11).

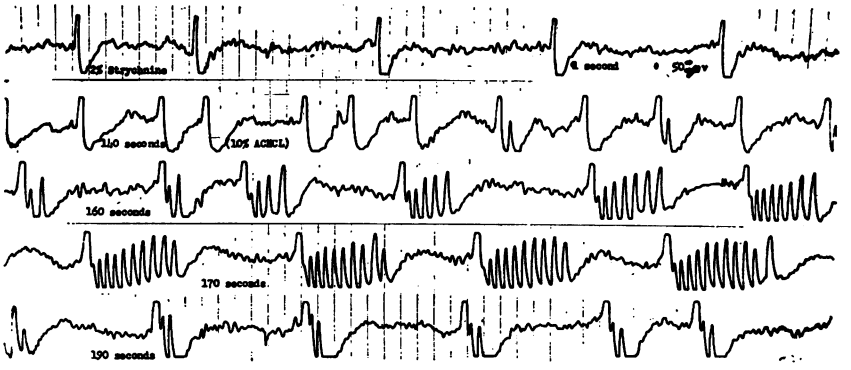

FIGURE 2—Serial electrocorticograms showing the local response of a strychninized right frontal cortical area after 10% acetylcholine chloride was applied for one minute (Case 1).

occurred in five of the nine times that the 10 per cent concentration was used and in three of the twenty times that the 20 per cent solution was employed.

There was usually a period of latency of six to eight minutes before significant electrocortical changes could be detected (Fig. 1). The first change noted was the presence of single negative waves of gradually increasing amplitude with a maximum of approximately 600 microvolts, and a frequency of one to two per second and a duration of 0.2 to 0.3 seconds. In the next phase of response, there occurred groups, clusters or spindles of increasing duration, in which high amplitude waves with a 5 to 8 per second frequency were dominant. With the 20 per cent acetylcholine chloride solution, the frequency within the spindles or groups was apt to center about 10 per second. The clusters, groups or spindles occurred in association with and immediately after the high amplitude single negative wave initially noted. In the ensuing phase, the initial high amplitude wave components were sometimes modified by the presence of an abrupt positive deflection so as to give the appearance of diphasic or triphasic spikes. Following the maximum response, the bursts of spindles, clusters or groups became less prominent, decreasing gradually in duration and amplitude. Thereafter the initial high amplitude negative waves and the spikes disappeared.

When a pledget of 10 per cent acetylcholine chloride solution was applied for one minute to a previously strychninized cortical area, a pronounced effect was noted (Fig. 2). Spikes immediately became very

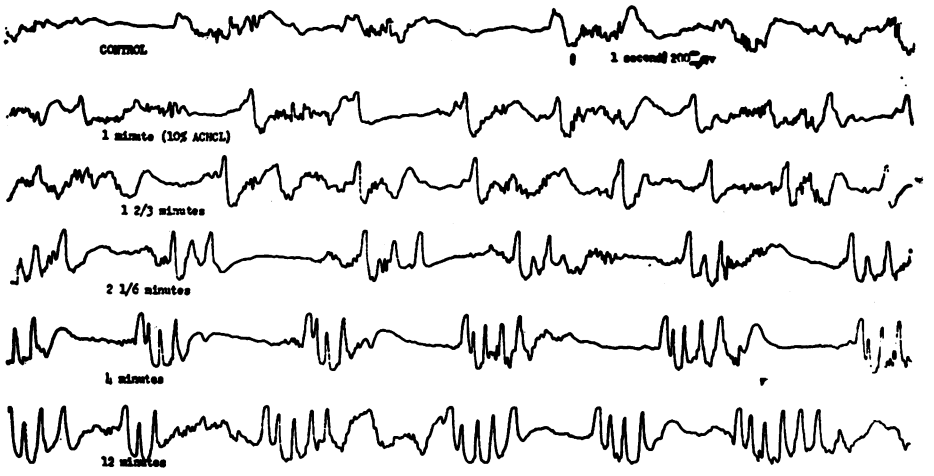

FIGURE 3—Serial electrocorticograms showing a decreased latency time and bursts of high amplitude 5 per second negative spikes following application of 10% acetylcholine chloride for twelve minutes to a previously responsive area (Case 2).

frequent and prominent and about one and a half minutes later high amplitude spindle bursts of 8 to 10 per second waves occurred immediately following the initial high amplitude negative spikes. This response reached its maximum three minutes after the acetylcholine chloride was applied and rapidly subsided thereafter in the ensuing one-half minute, the spindle bursts disappearing first. When a previously responsive cortical site was retested soon after the site had returned to its pre-test pattern, a greatly diminished latency in the onset of the new response sometimes occurred (Fig. 3).

Significant changes in the status of the patient and the cerebral cortex during and after the tests that could be attributed to the acetylcholine chloride were not noted.

Discussion: These findings indicate that significant alterations in the local electrocorticogram may occur several minutes after the topical application of 10 per cent or 20 per cent acetylcholine chloride solutions to human cerebral cortex. Although our series is small, there is a suggestion that the 10 per cent concentration may produce detectable changes more often than the 20 per cent concentration. Jordan, Badal and March[6] indicated that they obtained an "excitatory response" in 58 per cent of fifty-three applications of 10 per cent acetylcholine chloride in saline applied to the cat's cerebral cortex; our results with the use of

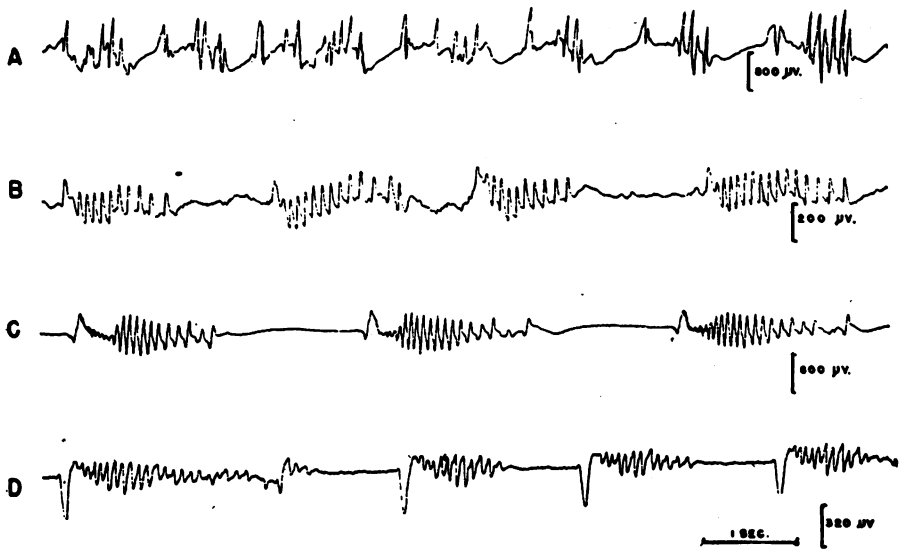

FIGURE 4—Representative electrocortical patterns following local application of 1% eserine and 0.5% acetylcholine to the cortex of the cat. A. Intact cortex; B. Thalamectomized hemisphere; C. Undercut cortex; 4. Isolated cortex. (Kristiansen, K. and Courtois, G. Electroencephalog. Clin. Neurophysiol. 1:265-71, 1949. Reproduced by permission.)

the 10 per cent solution (55 per cent) are in the same range. However, our overall results from use of both the 10 per cent and 20 per cent concentration are considerably lower (28 per cent).

We have not noted pronounced diminution of amplitude in the electrocorticogram in the period immediately after the test applications of acetylcholine chloride solutions. This is in contrast to some reports of the results in cats. Forster, Borkowski and McCarter[4] stated that in the cat, depression of electrical activity occurred locally within ten to forty seconds and then spread over the cortex in linear fashion, even in cases where no acetylcholine discharges occurred. This type of spreading depression encountered in the cat appears similar to that described by Leão[10] in the rabbit. Since Marshall and his workers[11-13] have indicated that factors such as dehydration and cooling are of extreme importance in the production of some spreading depressions, the absence of this finding in tracings made upon humans may be related to the vigorous efforts made to maintain appropriate hydration, temperature, electrolyte balance, etc. during surgical procedures in man. A species difference in reaction, possibly related to the comparatively higher

degree of cortical fissuration and the greater durability and complexity of the leptomeninges in man, may also be significant in this respect.

The maximum responses noted after the topical application of 10 per cent or 20 per cent acetylcholine chloride to human cerebral cortex resembled those described for animals after similar treatment with 10 per cent acetylcholine chloride or with eserine and 0.5 per cent concentration of acetylcholine chloride (Fig. 4). Facilitation of the acetylcholine chloride effect in man by previous strychninization is inferred from the prompt and pronounced response incurred under such circumstances. Forster and McCarter[5] noted that in cats strychninization of cortex followed by the application of acetylcholine chloride resulted in propagation of acetylcholine discharges to the area fired by strychnine, whereas ordinarily, acetylcholine discharges remained sharply localized to the region of application of the drug.

Since the average adult doses of atropine sulfate or scopolamine were routinely employed preoperatively, one would expect an adverse effect upon the response to a cholinergic substance such as acetylcholine chloride. However, it was felt that in most cases, the test period occurred after the presumed period of effectiveness of the preoperative medication. A distinct relationship of the response to the type and depth of anesthesia was not clearly demonstrated, and similar responses were noted under nitrous oxide, pentothal-nitrous oxide, avertin-nitrous oxide, and local anesthesia.

Significant cerebral pathological conditions were present in all cases, frequently in the area adjacent to the testing. In one instance (Case 8), however, a pronounced typical response occurred after the application of 20 per cent acetylcholine chloride solution to the cortex of a presumably intact cerebral hemisphere in a patient whose status was compatible with the clinical impression of a middle cerebral thrombosis of the opposite cerebral hemisphere. Altered leptomeningeal permeability was suggested as a factor especially in cases where gross physical changes in the pia were obvious.

A certain degree of control of the electrocortical response was possible by removal of the pledget from the cortex. Since we wished to avoid precipitation of frank motor convulsive phenomena, pledgets were always removed as soon as a relatively pronounced effect on the electrocorticogram was noted. Thereafter, the electrocortical response would begin to diminish in intensity and gradually disappear. An even

more pronounced electrocortical effect than any we have thus far encountered seems to be a definite probability in responsive cases through the use of more prolonged applications of the test substances. On some non-responsive occasions, however, the time or period of application appeared to be of minor importance; in one case 20 per cent acetylcholine chloride was applied continuously for sixty-three minutes without significant local electrocortical effect. The fact that some sites within a relatively small cortical area responded well and others did not, suggests the possibility of specific areal differences in the reaction to acetylcholine chloride. Since most of our test sites were within the frontal lobe, and because precise orientation was not always possible, an adequate basis for determining the presence or absence of a topographic pattern has not yet been attained.

Summary

1. Pronounced local changes in the human cerebral electrocorticogram could be induced by the topical application of acetylcholine chloride in 10 per cent or 20 per cent solutions.

2. In responsive cases, after a period of six to eight minutes, recurrent single, high amplitude one to two per second waves appeared.

3. Thereafter, recurrent spindles, groups or clusters of high amplitude 5 to 10 per second waves occurred in bursts immediately following the recurrent single high amplitude waves.

4. High amplitude diphasic and triphasic spike discharges were noted in the ensuing phase.

5. Recovery from the maximal response was characterized by the early diminution in duration and amplitude of the spindles, groups or clusters and was followed by the ultimate disappearance of the initial high amplitude negative waves and spikes.

6. Local early pronounced depression of electrical activity soon after the application of acetylcholine chloride solution was not noted.

7. Pretreatment of a cerebral cortical site with 2 per cent strychnine sulfate facilitated the response to 10 per cent acetylcholine chloride solution.

8. The influence of factors such as preoperative medication, anesthesia, cerebral pathological changes, leptomeningeal permeability, drug concentration, duration of application and site of application are considered.

(SEE REFERENCES ON FOLLOWING PAGE)

REFERENCES

1. Brenner, C. and Merritt, H. H. Effect of certain choline derivatives on electrical activity of the cortex, *Arch. Neurol. Psychiat. 48*:382-95, 1942.
2. Chatfield, P. O. and Dempsey, E. W. Some effects of prostigmine and acetylcholine on cortical potentials, *Amer. J. Physiol. 135*:633-40, 1942.
3. Forster, F. M. Action of acetylcholine on motor cortex; correlation of effects of acetylcholine and epilepsy, *Arch. Neurol. Psychiat. 54*:391-94, 1945.
4. Forster, F. M., Borkowski, W. J. and McCarter, R. H. Acetylcholine induced depression of cerebral cortical activity, *J. Neuropath. exp. Neurol. 5*:364-73, 1946.
5. Forster, F. M. and McCarter, R. H. Spread of acetylcholine induced electrical discharges of the cerebral cortex, *Amer. J. Physiol. 144*:168-73, 1945.
6. Jordan, W. K., Badal, D. W. and March, R. Interaction of effects of acetylcholine and adenosine triphosphate on electrical activity of cat cerebral cortex, *Arch. Neurol. Psychiat. 63*:766-73, 1950.
7. Kristiansen, K. and Courtois, G. Rhythmic electrical activity from isolated cerebral cortex. *Electroencephalog. clin. Neurophysiol. 1*:265-72, 1949.
8. Miller, F. R., Stavraky, C. W. and Woonton, G. A. Effects of eserine, acetylcholine and atropine on the electrocorticogram, *J. Neurophysiol. 3*:131-38, 1940.
9. Sjöstrand, T. Potential changes in the cerebral cortex of the rabbit arising from cellular activity and the transmission of impulses in the white matter, *J. Physiol. 90*:41*P*-43*P*, 1937.
10. Leão, A. A. P. Spreading depression of activity in the cerebral cortex, *J. Neurophysiol. 7*:359-90, 1944.
11. Marshall, W. H. The relation of dehydration of the brain to the spreading depression of Leão, *Electroencephalog. clin. Neurophysiol. 2*:177-85, 1950.
12. Marshall, W. H., Essig, C. F. and Dubroff, S. J. Essential factors involved in spreading cortical depression, *Amer. J. Physiol. 159*:579, 1949.
13. Marshall, W. H., Essig, C. F. and Dubroff, S. J. Relation of temperature of cerebral cortex to spreading depression of Leão, *J. Neurophysiol. 14*:153-66, 1951.

SUBSCRIPTION TO THE ACADEMY FOLDER

OCTOBER TO MAY 1952-53

The Folder of the Academy, listing meetings held during the academic year, will be mailed without request to all Fellows of the Academy residing within fifty miles of the Academy.

The Folder will also be mailed upon request and without charge to medical schools and hospitals within fifty miles of the Academy.

The Folder will be sent to other organizations and individuals at a subscription rate of $1.00 for the current academic year.